

HBeAg were co-incubated for 6h, compared to that of before stimulation, expression of B7-H1 on CD14⁺ cells increased, expression of TLR2 decreased, expression of B7-H1 and PD-1 on CD4⁺ and CD8⁺ T cells increased significantly, expression of costimulatory molecules CD28 on CD4⁺ cells decreased; Inflammatory factor TNF- α and anti-viral cytokine IFN- γ of HBeAg-stimulated PBMC from HBeAg-negative CHB patients, increased and reduced respectively.

Conclusion: By up-regulating expression of B7-H1 and PD-1, HBeAg may inhibit expression of TLR2 on CD14⁺ cells, reduce expression of costimulatory molecules CD28 on T lymphocyte, inhibit polarization of Th cells, and reduce active secretion of anti-viral factor of patients, which led to function of specific cell-mediated immunity became low, thereby clearance of virus by T lymphocyte were suppressed, which eventually resulted in persistence of HBV infection.

PP-108 Detecting hepatitis B virus large surface protein in patients with chronic hepatitis B: a clinical study

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Objective: To investigate the clinical significance of hepatitis B virus large protein (HBLP) detection in prediction of HBV DNA replication and the effect of anti-viral treatment.

Methods: The Serum samples were collected from 90 patients with HBV infection before and after anti-viral treatment. HBV DNA level was quantitatively detected using real-time polymerase chain reaction. HBLP was measured by enzyme-linked immunosorbent assay (ELISA). The collected data were analyzed by bivariate correlations method.

Results: There was no significant difference between the detectable rates of HBV DNA and HBV-LP in 90 blood samples before treatment ($p > 0.05$); The levels of serum HBV-LP was positively correlated with HBV DNA copies during anti-viral treatment ($r = 0.857$, $P = 0.000$). The OD value of HBV-LP and the copies of HBV-DNA decreased in the same trend along with treatment.

Conclusion: HBLP expression can reflect the state of HBV DNA replication. The decrease of HBLP and HBV-DNA during the anti-viral treatment could estimate the HBV replication state and predict the effect of anti-viral treatment.

PP-109 A study for HBsAg routine test negative results

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Objective: To see the false HBsAg negative results for serum samples in daily work and to improve the clinical laboratory tests quality.

Methods: Four hundreds HBsAg negative stochastic serum samples were collected from the copy tubes in daily work for detecting hepatitis B Virus markers (HBVM) with national ELISA reagent kits divided into 200 samples with HBsAb negative and 200 positive and put them -20° frostily. HBsAg markers were counterchecked with other two adding kinds of national reagent and American MONOLISA HBsAg ULTRA reagents (total 4 kinds) at the 400 samples and filtrated the positive samples. At the last filtrated samples, HBV DNA levels were doubly quantitative analyzed with fluorescence quantitative PCR (FQ-PCR) and taking the mean results.

Results: We deduced the conform HBsAg negative results from the three kinds of national reagents but five positive results from the American reagents in repeating HBsAg detection at the 200 HBsAb negative samples. No positive results were checked out from the 200 HBsAb positive samples with national or foreign reagents. The HBV DNA FQ-PCR quantitative results were all positive but less than 500 copies/ml.

Conclusion: The sensitive level of the HBsAg routine test ELISA

reagents is generally on the low side and easily bring on the false HBsAg negative results and the false results are more frequently from the HBsAb negative people. This maybe connected with occult HBV infection, we should attach importance to the HBVM counterchecking work at these people.

PP-110 Detecting hepatitis B virus large surface protein to filtrate the occult HBV infection

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Objective: Detecting hepatitis B virus large surface protein (HBLP) with serological method to filtrate the occult HBV infection and study the clinical detection strategy.

Methods: Two thousands HBsAg negative stochastic serum samples were collected from the copy tubes in daily work to detect hepatitis B Virus markers (HBVM) with national ELISA reagent kits and put them -20° frostily. The 2000 samples were detected with HBLP and filtrated the positive samples. HBsAg markers were doubly counterchecked with other two adding kinds of national ELISA reagent kits (total 3 kinds) at the filtrated samples. The last samples were doubly tested again with American MONOLISA HBsAg ULTRA reagents. HBV DNA levels were quantitative analyzed with fluorescence quantitative PCR (FQ-PCR) and taking the mean results.

Results: Fifteen HBLP positive samples were detected out from the 2000 serum samples. The conform negative results were deduced from the three kinds of national reagents but conform positive results from the American reagents in repeating HBsAg detection at the 15 samples. The HBV DNA FQ-PCR quantitative results were all positive but less than 500 copies/ml.

Conclusion: The false HBsAg negative results for serum samples are more generally from national reagents than from importations and HBLP results may be positive in these samples. Detecting HBLP marker is propitious to filtrate the occult HBV infection. This study provided a kind of serological reference for actively searching for the detecting strategy in occult HBV infection field.

PP-111 Evolution of hepatitis B virus in a chronic HBV-infected patient over 2 years

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Background: Evaluation of a 3-year evolution of hepatitis B virus in a chronically infected patient was conducted.

Methods: Clinic data and HBV DNA load(s) in serum were detected at three time points (1 day, 6 month and 31 month), HBV isolates were obtained at each time. The full-length HBV genome was cloned and 26 clones including three time points were randomly selected, sequenced and phylogenetically analyzed.

Results: All of the 26 clones belonged to subgenotype C2. 13 nucleotides in this subset of clones were found to be different from the published sequences of genotype C of HBV. During the 3-year evolution, nucleotides T361A, C930A, C2351T/A2353T, C2444T were the mutations been kept in and from minor to major while C339T and T770C were the new point mutations at the 3rd time point (31 month). Short fragment deletion (nt2849 to 2867) in the preS gene became dominant at the 31-month time point and had a trend to lead to large fragment deletion, which may produce presumptive P/S fusion proteins or truncated preS proteins. At the third time point, the patient's serum ALT, HBeAg and load of HBV DNA varied greatly.

Conclusion: The evolutionary data of HBV may provide clues for the interpretation of the course of HBV chronic infection and progressive pathology of liver diseases.

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PP-112 Analysis of five HBeAg-positive patients with Chinese herb Kuanxiongjiedu grain anti-HBV infection treatment

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Background and objectives: The objectives of the research are to observe the efficacy of the Chinese Herb Kuanxiongjiedu Grain (CHKG, consists of 12 ingredients) treatment for the patients whose HBV DNA levels over 10⁶ copies/mL and analysis the variations of the patients' hepatocyte.

Methods: Five patients with HBeAg-positive are selected. They have baseline ALTs: 22.3~285U/L, HBV DNAs: 2.1×10⁶~2.8×10⁸ copies/mL. The patients received CHKG two times daily (24g each time) for 32 weeks. The ALT and HBV DNA tests were implemented every two weeks in the first three months, and taken every four weeks in the following months. A Ultrasound Scanner was used to scan the patients and a normal person's livers. The mean and standard deviation of the gray levels of the normal person's scanned liver image are selected as the standard of normal liver.

Results: At the week 32, one patient's HBV DNA was below 1000 copies/mL and had normal ALT; three patient's HBV DNAs and ALTs have no significant changes; one patient's HBV DNAs have significant changes. Except the first patient, the patients' normal liver pixels increased 14%~51%.

Conclusion: The CHKG may help to recovered the chronic HBV patients' damaged hepatocyte even the patients' virus levels were not be suppressed during the therapy. Further investigation should be implemented.

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PP-113 Sequence analysis and replication fitness of the complete hepatitis B virus genome in patients with chronic hepatitis B

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Objective: To analyze the complete HBV genome in patients and evaluate its replication capacity.

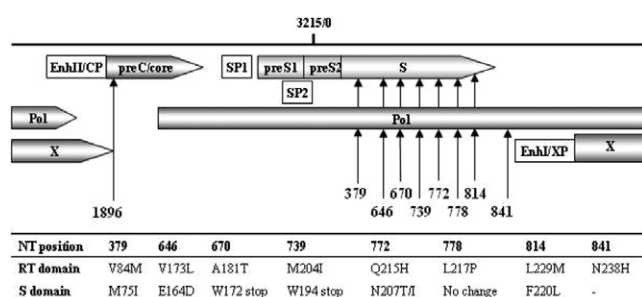


Fig. 1. Schematic diagram of the point mutations analyzed in the study. In the first part of the figure, the number on the top of the full HBV genome refers to the EcoRI site (3215/0). Position of the four open reading frames (ORFs) encoding preC/core, preS1/preS2/S, polymerase (Pol), and X protein are shown by the gray rectangle and the arrow indicates the transcription direction. The four cis-elements (enhancers and promoters) are indicated by the open triangle. The numbers under the full HBV genome represent the nucleotide positions of the studied mutations. In the second part of the figure are listed amino acid mutations in both polymerase (RT domain) and envelope genes-associated domains (S domain).

Methods: The full-length HBV genome amplification, cloning, and sequencing were preformed. Genotype and mutation sites related to antiviral agents were analyzed, and site-directed mutagenesis was performed on interested sites. The full-length HBV genomes were transfected into HepG2 and Huh-7 cell lines. 72 h after transfection, expression of HBsAg was detected with ELISA, quantitation of intracellular HBV replicative intermediates were examined by qPCR.

Results: 12 different clones were obtained. several mutation sites, such as A181V/S, V84M, were identified, and the better management of 5 patients was developed.

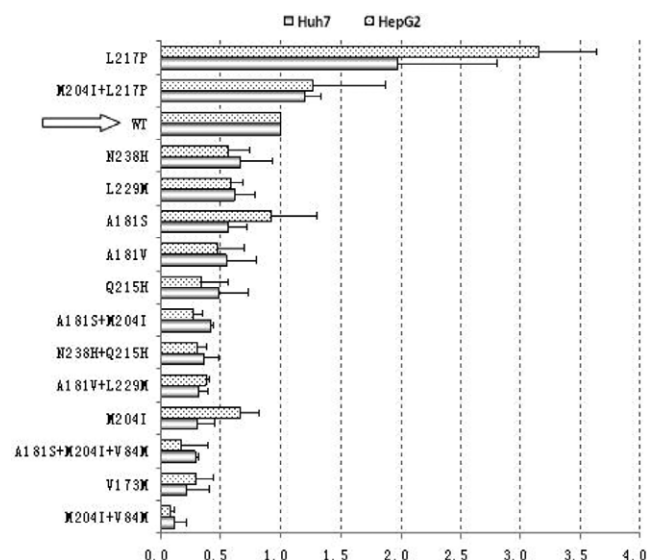


Fig. 2. Relative replication yield of HBV mutants.

Conclusion: Vector-free replication assay is an approach to determine the phenotype of clinical HBV strains, which could become an important tool for the management of patient infected with HBV.

PP-114 Correlation factors involved in therapeutic efficacy of adefovir dipivoxil for chronic hepatitis B with YMDD mutation

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Background: To investigate the correlation factors associated with the therapeutic efficacy of adefovir dipivoxil for chronic hepatitis B with YMDD mutation.

Methods: 92 patients were enrolled and treated with adefovir dipivoxil for 48 weeks, Logistic regression analysis was used to identify some possible correlation factors associated with therapeutic efficacy.

Results: Patients who achieved undetectable HBV DNA at week 48 of the treatment were found to have a lower baseline HBV DNA levels compared with those who did not achieve, and the same result in patients who achieved HBeAg seroconversion and serum alanine aminotransferase (ALT) normalization. There were significant difference between patients whose baseline ALT levels ≤1 ULN, HBV DNA levels ≤5.0 lgcopies/mL and patients whose baseline serum ALT levels >1 ULN, HBV DNA levels >5.0 lgcopies/mL in undetectable HBV DNA ($\chi^2=17.321$, $P<0.001$), HBeAg seroconversion ($\chi^2=3.88$, $P=0.049$) and ALT normalization rates ($\chi^2=25.526$, $P<0.001$) after 48-week treatment. Logistic regression analysis indicated the baseline HBV DNA levels, undetectable serum HBV DNA by PCR at week 24, and undetectable serum YMDD mutation at week 12 were correlation factors of therapeutic efficacy at week 48.

Conclusion: Better response at week 48 has significantly asso-